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June 1982

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THE EFFECTS OF MICROWAVES ON ANIMAL OPERANT BEHAVIOR

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MF58.524.02C-0010

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25 June 1982

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SUMMARY PAGE

THE PROBLEM

Fleet personnel are frequently exposed to microwave irradiation produced by various weapons systems and communication devices. Microwave irradiation has been shown to cause behavioral effects in non-human animals and may cause behavioral changes in humans. In order better to generalize from animals to humans several different sized animals of different species should be used as subjects. This report summarizes the findings of behavioral experiments that are part of an interdisciplinary effort whose final goal is to document the biological effects of microwaves on animals, extrapolate these to man, and then propose exposure standards for fleet personnel.

FINDINGS

✓ Rats, squirrel monkeys, and rhesus monkeys trained to respond on operant behavioral tasks and then exposed to microwaves produce data suggestive of a possible extrapolation to humans in similar situations. The data indicate that the behavioral effects are dependent upon an animal's reaction to thermogenous energy. Even when an animal's performance on an operant task involved relearning the task daily, the disruption of performance was still dependent upon the animal's reaction to total absorbed microwave induced heat and/or the distribution of that heat.

ACKNOWLEDGMENTS

The collaboration with J. Knepton on the repeated acquisition portion of this paper and the continued assistance of C. S. Ezell are gratefully acknowledged. Especial appreciation is extended to Mrs. Roberts for her assistance in typing the manuscript.



The experiments reported herein were conducted according to the principles set forth in the "Guide for the Care and Use of Laboratory Animals," Institute of Laboratory Animal Resources, National Research Council, DHEW, PUB. No. (NIH) 78-23.

INTRODUCTION

The nature of the biological effect of microwaves upon various animals may be elusive when one considers extremely low levels of microwave energy, but in terms of hazard the only well documented effect is that caused by the thermal characteristics of deposited microwave energy. On the basis of the thermal hypothesis, several broad areas of research can be identified which might allow one to correlate findings in different disciplines utilizing the same animal species, exposure chambers, and microwave parameters. Once such relationships are explored it should be possible to not only extrapolate from the various animal models, but to also establish thresholds for safe exposure of humans to microwaves. The Bioenvironmental Sciences Department at the Naval Aerospace Medical Research Laboratory, Pensacola, Florida, has embarked on such an endeavor, and this report summarizes a series of experiments in which the behavior of animals undergoing microwave exposure is studied as a fundamental part of the project (1-4, 7, 10). Concurrently, research on endocrinology and metabolism, epidemiological surveys of personnel and their microwave environment, dosimetric studies in rat, monkey, and human models, and basic research on the biophysics of interaction mechanisms are being conducted.

PROCEDURE

SUBJECTS

Rats, typically of the long Evans strain obtained from the Charles River colonies and normally weighing in the range of 300-400 g, were used as subjects as were squirrel monkeys, Saimiri sciureus, obtained from Columbia, South America. In addition, rhesus monkeys, Macaca mulatta, bred in our own laboratory were used. The general body mass of the squirrel monkeys was approximately 700 g and that of the rhesus was approximately 5 kg. In the behavioral studies the animals were generally deprived of food and maintained at approximately 85% of their free-feeding body mass. These specific species were chosen for various reasons including their being representative of mammals, their unique body-size relationships to each other (almost logarithmic), and their cranial cavities having diameters roughly the size of one of three microwave wavelengths (multiplied by 0.4) used to irradiate the animals. In addition, the rhesus monkey was chosen as a subject because its heat-handling ability and distribution of temperature sensitivity over the body are similar to those of man.

APPARATUS

All microwave exposures occurred in anechoic chambers which differed only in basic dimensions, dependent upon the frequency of the radar used to irradiate the animals or models. Each chamber was constructed of copper-shielded wood and lined with pyramidal absorber. The chambers were cooled with air conditioners and ventilation fans, and each was equipped with a closed circuit television camera. Equipment for presenting either visual or auditory stimuli was in the chambers during the behavioral studies. The chambers also included devices for recording ambient humidity and temperature and for fire detection and extinction.

Various military and commercial radar sets were used to generate the microwaves at three different frequencies: 1.28, 2.45, and 5.62 GHz. The 2.45 GHz microwaves were 100% amplitude (sinusoidally) modulated at 120 Hz, whereas the other frequencies were pulsed (370 pps with a pulse duration of 3 μ s at 1.28 GHz, 662 pps with a pulse duration of 2.0 μ s at 5.62 GHz). In most cases the animals were irradiated in the far-field of the horn with frontal, vertically polarized irradiation; in some experiments at 2.45 GHz irradiation was from above. Incident power density and its distribution in the chamber were determined in each experiment without the subject, and that power density corresponding to the center of a subject's head was the value referred to as the one illuminating the animal.

Unique to this research project was the development of Styrofoam restraint devices (9). All of these experiments used variations of such devices either as restraint chairs, animal conditioning boxes, or molds for models while irradiation occurred. In the behavioral experiments these Styrofoam devices also contained various manipulanda that the animals operated to obtain food. In the case of models, negative images were either carved or molded in Styrofoam and filled with physiological saline or flesh-simulating material (8).

In the experiments with monkeys, colonic temperatures were obtained during irradiation; in the rat studies, colonic temperatures were normally obtained immediately before and after irradiation. Reference temperature probes were located near the colonic probes but outside of the animals.

The anechoic chambers provided some sound attenuation. In addition, a masking noise of approximately 75 dB was delivered to the chambers during behavioral studies.

METHODS

Full-scale models of rats, rhesus monkeys, and man have been produced by R. G. Olsen in our laboratory. The sitting rhesus model of flesh-simulating material has been exposed to 1.29 GHz, and both calorimetric and thermographic determinations of averaged specific absorption rate (SAR) have been made (8).

In studies by W. G. Lotz, rhesus monkeys in restraint chairs were instrumented with indwelling jugular catheters and exposed to 1.29-GHz microwaves for 8 hours during the day and also at night. The monkeys were repeatedly exposed over a period of one week. Blood samples were taken every hour during 24-hour periods, and plasma cortisol and plasma thyroxine levels along with colonic temperature were determined. Power densities of 20, 28, and 38 mW/cm² were used (6). Rats were exposed to 1.29 and 2.45 GHz microwaves for 2 hours, and plasma corticosterone was determined by sampling from indwelling catheters in the jugular vein.

The standard methodology in the behavioral experiments has been to train the animals in hard plastic, Plexiglas, cages until stable performance on an operantly conditioned task develops. The animal was then transferred to a Styrofoam container, stable performance was recovered, and then the animal was irradiated. A period of 3 to 12 months elapsed during this

period, depending upon the species used. Rats involved less time than rhesus monkeys. The animals were handled frequently and learned to enter their restraint devices without obvious signs of stress.

Most of the operant tasks utilized in our approach have had food-deprived animals responding on one of two different levers to produce a small food pellet. Signals were presented randomly, dependent upon an animal's response rate, and when the signal corresponding to food appeared, the animal had to make a response on a different lever to obtain the food. Most of their daily food allotment was obtained performing this task. Variations in the frequency of reinforcement, the number of manipulanda, and the number and modality of stimuli have occurred throughout the length of our investigations. Food was the only reinforcer used and water was not available during the experimental sessions which have lasted from 40 minutes to 2 hours. The primary measure of performance has been the rate of responding on a lever, but other measures of performance such as reaction time, errors of omission and commission, and failure to respond have also been obtained.

Our most recent behavioral work involved squirrel monkeys trained to respond on one of three specific levers in the presence of one of three specific auditory and visual stimuli. The animal learns the task in one session, and the lever-stimuli relationship is changed for the next session. Session-to-session changes in these stimulus-response arrangements generate daily learning curves. These curves become relatively stable over several sessions, thus allowing one to investigate not only ongoing performance, but also the acquisition of behavior. We have completed two studies in this area, one in which the animal was first irradiated and then performed the task outside of the irradiation chamber (7), and the other in which the animal performed the task in a restraint chair (Fig. 1) during irradiation (unpublished experiment by J. Knepton).

In all experiments behavior was allowed to reach session-to-session stability as determined by response-rate equivalency and then the animals were irradiated. Typically each irradiation session was preceded and followed by sham sessions in which everything was the same except the magnetron was not energized. A threshold, in terms of power density value, was determined by assessing the level at which stable performance was disrupted. This disruption generally took the form of a decrease in response rate on the appropriate lever. Simultaneously, when possible, colonic temperatures were determined corresponding to that threshold.

RESULTS AND DISCUSSION

Thermographic analysis of the sitting rhesus revealed head, arm, and leg resonances at 1.29 GHz. These findings were confirmed with a Seebeck-type gradient-layer calorimeter. The largest amount of heating occurred in the legs and knees of the sitting rhesus model (8).

Temperature measurements in the rat and squirrel monkey models, obtained with a Vittek-Model 101 Electrothermia Monitor, also revealed unique distributions of absorbed energy. The distribution of energy in the rat model was dependent upon microwave frequency. The 1.29-GHz microwaves

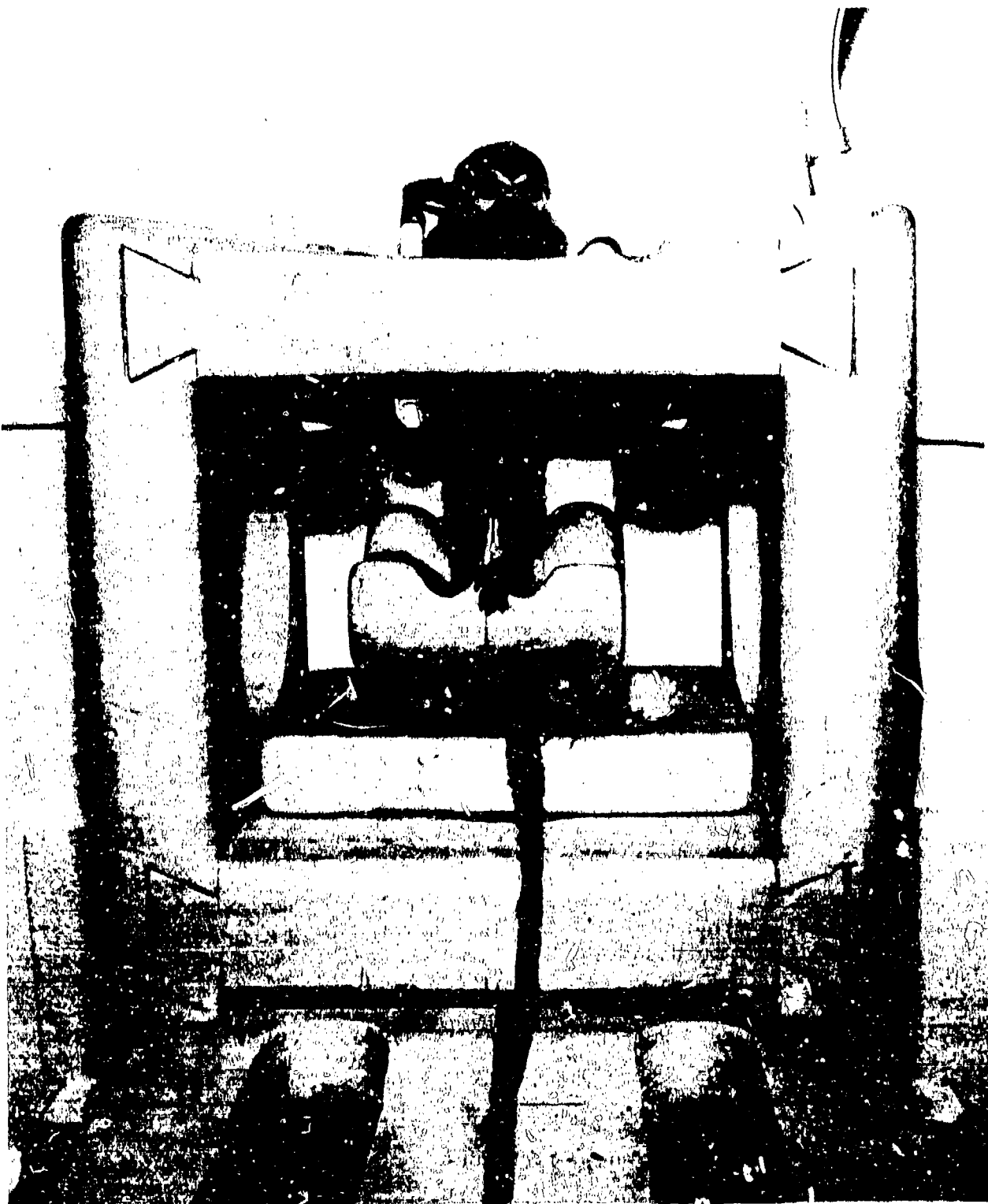


Figure 1

A squirrel monkey seated in a restraint chair for the study of repeated acquisition. Three levers protrude through the top surface of the chair and the food delivery tube is on the right of the chair.

deposited relatively larger amounts of energy near the far surface of the model, and the 5.62-GHz microwaves deposited greater amounts of energy at the irradiated surface. The measurements in the squirrel monkey model at 5.62 GHz also revealed relatively greater amounts of deposited energy on the front surface, arms, legs, and tail. Virtually no energy penetrated to the rear surface of the squirrel monkey model at 5.62 GHz.

The microwave exposures of rats at 1.29 GHz resulted in significant increases in circulating corticosterone level at lower amounts of power density (15 mW/cm²) than needed at 2.45-GHz exposures. Power densities of 20-30 mW/cm² were needed at 2.45-GHz exposures to cause similar changes in corticosterone level. This finding is probably related to the energy distribution differences revealed in the dosimetry analysis (6).

Exposures of the rhesus monkeys to 1.29-GHz irradiation also produced increases in the adrenocortical response. The threshold for an increase in circulating cortisol level was approximately 38 mW/cm² for 8-hour exposures (from 1200 to 2000 hours). Lotz' work also found this response to be dependent upon the circadian rhythm. Although similar rectal temperature increases were observed for both day and night exposures (1.7 and 1.8 °C, respectively), night exposures (between 2200-0600 hours) did not result in similar cortisol increases.

Another interesting aspect of colonic temperature measurements while the monkeys were being exposed was the fact that virtually all animals showed no behavioral effect until their temperatures increased by 1 °C or more. This effect has not yet been confirmed at frequencies other than 2.45 GHz. Other observations involving temperature measurements in the monkeys revealed that colonic temperature increased (approximately 0.5 °C in 120 minutes) throughout an experimental session even during sham exposures and when an animal stopped lever responding, rectal temperature stopped increasing and often decreased.

With repeated exposures of the animals to microwaves at various power densities between 0 and 75 mW/cm², it was possible to determine the average power density in each animal where disruption of behavior occurred. The medians of these various power densities were then used to calculate thresholds of disruption in each of the studies. The thresholds (Fig. 2) for animals in 60-minute exposures at 2.45 GHz are 28 mW/cm² for rats, 45 mW/cm² for squirrel monkeys, and 67 mW/cm² for rhesus monkeys. Similarly, for rats in 40-minute exposures to 1.28 and 5.62 GHz, the thresholds for behavioral disruption are approximately 10 mW/cm² and 26 mW/cm², respectively. The experiments with rhesus and squirrel monkeys exposed to 1.28- and 5.62-GHz microwaves are not complete, but preliminary data reveal that rhesus monkeys do not have their behavior measurably disturbed at levels less than 40 mW/cm² when exposed to 1.28-GHz microwaves for 60 minutes or less.

The effect with squirrel monkeys working a repeated acquisition task was seen between 40 and 50 mW/cm² if the animals undergo 60 minutes of 5.62-GHz exposures before working the task; and between 38 and 46 mW/cm² if exposed for 60 minutes while working the task. The difference between these values is probably of no practical consequence, and the value of 45 mW/cm², which corresponds to the value obtained at 2.45 GHz, is a relatively

good estimate for behavioral disruption on this task also.

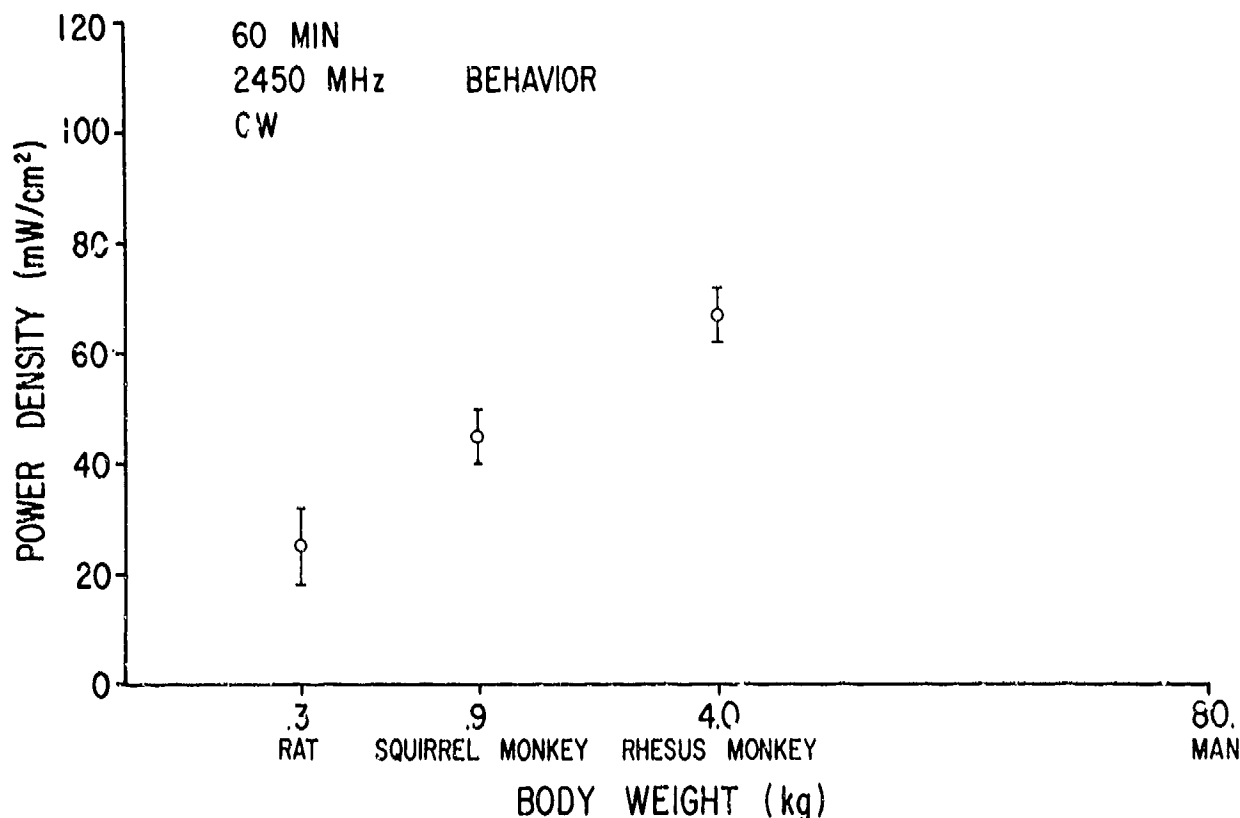


Figure 2

The average minimal power densities corresponding to behavioral disruption on operant tasks are plotted as a function of body weight.

With a finer examination of the repeated acquisition data, several consistencies appear in both the behavioral response and in colonic temperature (Fig. 3). The correct responses (normalized as a percentage of total responses per food pellet delivered) during irradiated sessions did not differ from correct responses during sham sessions until the power density reached 38 mW/cm² (mean of four monkeys). This difference was concentrated in the final 30 minutes of a 60-minute session during which the monkeys made substantially fewer correct responses while undergoing microwave exposure. Note, however, that under both sham and irradiated conditions, approximately the same percentage of correct responses occurred prior to a deterioration of performance. This observation indicates to the present investigator that learning, per se, was not affected at this power density; only the performance of the already learned task was disrupted by the microwaves. Even at 46 mW/cm², exposed monkeys displayed a learning curve; however, peak performance under irradiated conditions never reached the

same level as under sham conditions (55% correct responses versus 70%). Nevertheless, the learning curve still increased during the initial 40 minutes of the session; hence, this power density of microwave irradiation certainly did not prevent the occurrence of learning.

Colonic temperature during the repeated acquisition task always increased even in the absence of irradiation, and part of this increase is attributed to the metabolic heat generated while the animal manipulated the levers. When the task terminated and the levers were no longer functional, colonic temperature tended to decrease slightly. Ambient room temperature was held constant during these sessions. Under every power density colonic temperature increased regardless of whether behavioral changes occurred. The increase in colonic temperature was correlated with the increase in power density as observed in all of our primate experiments.

CONCLUSIONS

The research approach of this investigator regarding biological effects of acute exposure to microwaves has been to examine behavior not only as a correlate of many physiological processes but also as the final common reflection of those processes. Hence, behavior should reflect dramatic acute environmental insult in addition to compensating homeostatically for that insult. Under these circumstances, only when behavior is disrupted by a specific level of microwave irradiation is that level considered hazardous. If performance is improved by irradiation, those conditions of irradiation are not considered to be hazardous to the organism. Some investigations have demonstrated changes in behavior as a consequence of irradiation such as increases or decreases in general motor activity, but the direction of these changes has not been defined in such a manner as to permit a definite assessment of hazard.

The behavioral research in our laboratory has not uncovered any behavioral effects unrelated to heat production by microwaves in the organism. In virtually all cases where behavior has been disrupted substantial increases in colonic temperature also occurred. Normally these increases have been on the order of 1 ° to 2 °C above sham irradiation, but it should be noted that assessment of baseline temperatures has to take into consideration habituation to handling and restraint and also temperature differences dependent upon the animal's circadian rhythm.

Our dosimetry has also revealed that the behavioral changes we have observed may be dependent upon other than whole-body heating. Even heating of the central nervous system may not be the culprit. The possibilities exist that visceral heating may serve to decrease motivation or heating in a limb serve as a distracting stimulus. All of us are familiar with the microwave auditory phenomenon, but investigations as to other local thermal phenomena in the absence of whole-body heating have not been forthcoming.

On the other hand, our research in which whole-body heating was measured has provided minimal information for speculation as to safe levels of microwave exposure for humans (12) and the foundation for future research by other investigators (11).

Our work has also revealed that even in cases where incident power density at one microwave frequency is the same as at a different microwave frequency, differential amounts of energy may be deposited in an animal, resulting in different behavioral effects. The distribution differences in thermal energy are also responsible in this case even though specific hot spots may not develop.

The only cumulative effects found in our behavioral studies were those instances where animal behavior was disrupted less with repeated exposures to irradiation. Such habituation occurred numerous times, not only as revealed in behavior but also in colonic temperature. Habituation to repeated presentation of microwaves occurred at all three frequencies used (1.28, 2.45, and 5.62 GHz).

In conclusion, our program has so far provided the initial steps in establishing a format for a comprehensive examination of the hazards of microwave exposure. Although future work may reveal some inadequacies in this approach, it is certain that similar parametric studies have to be conducted in a similar interdisciplinary manner.

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REPORT DOCUMENTATION PAGE		READ INSTRUCTIONS BEFORE COMPLETING FORM
1. REPORT NUMBER NAMRL - 1285	2. GOVT ACCESSION NO. AD-A127486	3. RECIPIENT'S CATALOG NUMBER
4. TITLE (and Subtitle) The Effects of Microwaves on Animal Operant Behavior		5. TYPE OF REPORT & PERIOD COVERED
7. AUTHOR(s) John O. de Lorge		6. PERFORMING ORG. REPORT NUMBER
9. PERFORMING ORGANIZATION NAME AND ADDRESS Naval Aerospace Medical Research Laboratory Naval Air Station Pensacola, Florida 32508		10. PROGRAM ELEMENT, PROJECT, TASK AREA & WORK UNIT NUMBERS MF58.524.02C-0010
11. CONTROLLING OFFICE NAME AND ADDRESS Naval Medical Research and Development Command National Naval Medical Center Bethesda, Maryland 20014		12. REPORT DATE 25 June 1982
14. MONITORING AGENCY NAME & ADDRESS (if different from Controlling Office)		13. NUMBER OF PAGES 13
		15. SECURITY CLASS. (of this report) Unclassified
		15a. DECLASSIFICATION/DOWNGRADING SCHEDULE
16. DISTRIBUTION STATEMENT (of this Report) Approved for public release; distribution unlimited.		
17. DISTRIBUTION STATEMENT (of the abstract entered in Block 20, if different from Report)		
18. SUPPLEMENTARY NOTES		
19. KEY WORDS (Continue on reverse side if necessary and identify by block number) Operant Behavior Microwaves and behavior rats squirrel monkeys rhesus monkeys		
20. ABSTRACT (Continue on reverse side if necessary and identify by block number) Acute effects of microwave irradiation on behavior were investigated as part of an interdisciplinary research effort. Other areas of concern were measurements of absorbed energy and its distribution, endocrinology and measurement of environmental exposures along with an epidemiological survey. The final goal of these research endeavors is to eventually discover thresholds for human nonhazardous exposure to microwaves. Rats, squirrel monkeys, and rhesus monkeys trained on operant behavioral tasks and exposed to microwaves		

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